Socioeconomic Status and the Risk of Cervical Intraepithelial Neoplasia Grade 3 among Oncogenic Human Papillomavirus DNA-Positive Women with Equivocal or Mildly Abnormal Cytology

Michelle J. Khan, B.A.[1,2]
Edward E. Partridge, M.D.[3¶]
Sophia S. Wang, Ph.D.[1]
Mark Schiffman, M.D., M.P.H.[1]
for the Atypical Squamous Cells of Undetermined Significance/Low-Grade Squamous Intraepithelial Lesion Triage Study (ALTS) Group

[1] Division of Cancer Epidemiology and Genetics, National Cancer Institute, National Institutes of Health, Department of Health and Human Services, Rockville Maryland.

[2] HHMI–NIH Research Scholars Program, Howard Hughes Medical Institute, Chevy Chase, Maryland.

[3] Department of Obstetrics and Gynecology, University of Alabama at Birmingham, Birmingham, Alabama.

Supported by the National Cancer Institute, National Institutes of Health, Department of Health and Human Services contracts CN-55153, CN-55154, CN-55155, CN-55156, CN-55157, CN-55158, CN-55159, and CN-55105.

Some of the equipment and supplies used in this study were donated or provided at reduced cost by Digene Corporation (Gaithersburg, MD), Cytyc Corporation (Boxborough, MA), National Testing Laboratories (Fenton, MO), Denvu (Tucson, AZ), TriPath Imaging, Inc. (Burlington, NC), and Roche Molecular Systems Inc. (Alameda, CA).

The Atypical Squamous Cells of Undetermined Significance (ASCUS)-Low Grade Squamous Intraepithelial Lesion (LSIL) Triage Study (ALTS) Group Coauthors: National Cancer Institute, Bethesda, MD (D. Solomon, Project Officer; M. Schiffman, Coproject Officer; R. Tarone, Statistician); ALTS Clinical Centers: University of Alabama at Birmingham, Birmingham, AL (E. E. Partridge, Principal Investigator; L. Kilgore, Coprincipal Investigator; S. Hester, Study Manager); University of Oklahoma, Oklahoma City, OK (J. L. Walker, Principal Investigator; G. A. Johnson, Coprincipal Investigator; A. Yadack, Study Manager); Magee-Womens Hospital of the University of Pittsburgh Medical Center Health System, Pittsburgh, PA (R. S. Guido, Principal Investigator; K. McIntyre-Seltman, Coprincipal Investigator; R. P. Edwards, Investigator; J. Gruss, Study Manager); University of Washington, Seattle, WA (N. B. Kiviat, Coprincipal Investigator; L. Koutsky, Coprincipal Investigator; C. Mao, Investigator); Colposcopy Quality Control Group (D. Ferris, Principal Investigator, Medical College of Georgia, Augusta, GA; J. T. Cox, Coinvestigator, University of California at Santa Barbara, Santa Barbara, CA; L. Burke, Coinvestigator, Beth Israel Deaconess Medical Center Hospital, Boston, MA); HPV Quality Control Group (C. M. Wheeler, Principal Investigator, University of New Mexico Health Sciences Center, Albuquerque, NM; C. Peyton-Goodall, Laboratory Manager, University of New Mexico Health Sciences Center, Albuquerque, NM; M. M. Manos, Coinvestigator, Kaiser Permanente, Oakland, CA); Pathology Quality Control Group (R J Kurman, Principal Investigator, Johns Hopkins Hospital, Baltimore, MD; D. L. Rosenthal, Coinvestigator, Johns Hopkins Hospital, Baltimore, MD; M. E. Sherman, Coinvestigator, The National Cancer Institute, Rockville, MD; M. H. Stoler, Coinvestigator, University of Virginia Health Science Center, Charlottesville, VA); Westat, Coordinating Unit, Rockville, MD (J. Rosenthal, Project Director; M. Dunn, Data Management Team Leader; J. Quarantillo, Senior Systems Analyst; D. Robinson, Clinical Center Coordinator); Quality of Life Group (D. M. Harper, Chair of ALTS Quality of Life Group, Dartmouth Medical School, NH); Digene Corporation, Gaithersburg, MD (A. T. Lorincz, Senior Scientific Officer); and Information Management Services, Inc., Silver Spring, MD (B. Kramer, Senior Programmer/Analyst).

Address for reprints: Edward E. Partridge, M.D., Department of Obstetrics and Gynecology, University of Alabama at Birmingham, 618 South 20th Street, Birmingham, AL 35233; Fax: (205) 975-6174; E-mail: eep3840@uab.edu

Received August 23, 2004; revision received February 2, 2005; accepted February 23, 2005.

BACKGROUND. Low socioeconomic status (SES) is a reported risk factor for cervical carcinoma, but few studies have taken into account adequately the possibly confounding effects of oncogenic human papillomavirus (HPV) infection as well as access to screening and subsequent treatment.

METHODS. Women (n = 5060 women) with a mean age of 27.5 years and with equivocal or mild cytologic cervical abnormalities were enrolled in the Atypical Squamous Cells of Undetermined Significance/Low-Grade Squamous Intraepithelial Lesion (ASCUS-LSIL) Triage Study (ALTS), a clinical trial that evaluated management strategies. The women were seen every 6 months for 2 years. The enrollment questionnaire assessed three indicators of SES: race/ethnicity, education, and source of payment for medical care. Multivariate logistic regression models were used to identify predictors of oncogenic HPV DNA positivity at enrollment and to assess associations between the SES indicators and risk of cervical intraepithelial neoplasia grade 3 (precancer) and carcinoma (≥ CIN3) identified

Published 2005 by the American Cancer Society*
DOI 10.1002/cncr.21129
Published online 11 May 2005 in Wiley InterScience (www.interscience.wiley.com).

throughout the study (n = 506 women) among oncogenic HPV-positive women (n = 3133 women).

RESULTS. SES indicators were not associated significantly with oncogenic HPV infection after adjustment for age at enrollment, recent and lifetime number of sexual partners, study center, and smoking history. Among women with oncogenic HPV, the risk of ≥ CIN3 increased with decreasing education (less than high school education: odds ratio [OR], 2.4; 95% confidence interval [95%CI], 1.5–3.7 vs. completed college). Black women (OR, 0.5; 95%CI, 0.4–0.7) and white/Hispanic women (OR, 0.4; 95%CI, 0.2–0.8) were at decreased risk for ≥ CIN3 compared with white/non-Hispanic women. The source of payment for medical care was not associated with risk.

CONCLUSIONS. Factors associated with lower SES, such as low education, may serve as a surrogate for unknown factors that influence progression to ≥ CIN3 among women with oncogenic HPV infection. In this controlled setting with equalized follow-up and treatment, the decreased risk of ≥ CIN3 associated with black and white/Hispanic race/ethnicity could be further examined. Ongoing efforts should emphasize methods for equalizing screening and follow-up among women of varying SES, regardless of race or ethnicity. ***Cancer*** **2005;104:61–70.**

KEYWORDS: cervical intraepithelial neoplasia, socioeconomic status, human papillomavirus, human papillomavirus, triage, cofactors.

Infections by ≈ 15 oncogenic human papillomavirus (HPV) types cause virtually all cases of cervical carcinoma, which is the second most common malignancy in women worldwide.[1] Most women clear HPV infections, with or without accompanying HPV-induced cytologic abnormalities, within 2 years. Uncommonly, some oncogenic HPV infections persist, and women with persistent infection have an elevated risk of developing cervical intraepithelial neoplasia Grade 3 (CIN3), the immediate precursor to carcinoma, and cervical carcinoma.

Multiple epidemiologic studies have identified secondary risk factors (HPV cofactors) that are associated with the development of CIN3 or carcinoma (≥ CIN3) from oncogenic HPV infection, including long-duration oral contraceptive (OC) use,[2–4] multiparity,[3,5] smoking,[3,6–8] host immune function,[9] and non-HPV sexually transmitted infections.[10–13] For this article, we examined socioeconomic status (SES) as a possible cofactor for HPV.

Several studies have reported an inverse association between SES indicators and invasive cervical carcinoma.[14,15] SES usually is measured by indicators such as education and income; however, in the United States, race also can be considered a proxy for SES. Increased risks of cervical carcinoma incidence and mortality among women of black and Hispanic race, compared with women of non-Hispanic white race, have been reported.[15–19] Asian race, to a lesser extent, also has been associated with increased risk of cervical carcinoma.[20] Although it has been suggested that health disparities may be due to biologic differences between individuals of various races, current scientific evidence supports that health outcomes often are equal if equal screening and treatment are given.[21–23] Nevertheless, the issue is not solved, because racial/ethnic disparities in health outcomes sometimes remain, even after accounting for medical insurance status, income, age, and severity of conditions.[24]

Based on international correlation studies,[25] we postulated that any association of SES or race/ethnicity with cervical carcinoma incidence mainly may be a consequence of two factors: 1) differences in infection by human papillomavirus (HPV), the causative agent of cervical carcinoma, and/or (2) differential screening and follow-up. In the current study, we sought to determine whether SES remained an independent risk factor for cervical neoplasia after considering HPV infection and access to screening/follow-up. Analyses were conducted within a screened population of women who were enrolled as part of the Atypical Squamous Cells of Undetermined Significance (ASCUS)-Low Grade Squamous Intraepithelial Lesion (LSIL) Triage Study (ALTS), a 5060-woman, randomized trial that was conducted by the United States National Cancer Institute.

MATERIALS AND METHODS

Study Design and Population

ALTS was a randomized trial conducted by the National Cancer Institute (NCI) (National Institutes of Health, Rockville, MD) comparing three triage strate-

gies for women with ASCUS or LSIL; details of the design, methods, and primary results of ALTS have been published elsewhere.[26–30] Briefly, women with ASCUS or LSIL cytology were recruited to participate in the study at four clinical centers: the University of Alabama at Birmingham (Birmingham, AL), Magee-Womens Hospital of the University of Pittsburgh Medical Center Health System (Pittsburgh, PA), the Oklahoma University Health Sciences Center (Oklahoma City, OK), and the University of Washington (Seattle, WA). NCI and local institutional review boards approved the study. In total, 5060 women enrolled in the study from January, 1997 to December 1998: 3488 women with ASCUS and 1572 with LSIL cytology. Routine follow-up and exit visits concluded in January 2001.

Questionnaire Data

Each ALTS participant was administered a questionnaire at enrollment to collect information on demographics, lifestyle, and medical history, as described previously.[26] The enrollment questionnaire included medical, reproductive/contraceptive, gynecologic, sexual, demographic, and smoking history. The demographic variables that we considered SES indicators for this study included race/ethnicity, education, and source of payment for medical care (abbreviated here as "source of medical care"). Ethnicity was self-reported as Hispanic or non-Hispanic, and race was self-reported as white, black, Asian/Pacific Islander, or American Indian/Alaskan Native. For simplicity, we condensed these two variables into a single "race/ethnicity" variable with five categories: white/non-Hispanic, white/Hispanic, black, Asian/Pacific Islander, and American Indian/Alaskan Native. Education was self-reported in eight categories, which we condensed into four: less than high school, completed high school, some college/vocational school, and completed college. Source of medical care was self-reported in six categories and was recategorized into four: no source, Medicaid/Medicare/other government assistance, self-pay/family pay, and insurance.

HPV DNA Testing

Hybrid Capture 2 (Digene Corporation, Gaithersburg, MD) using the probe set B (henceforth referred to as HC2) is a DNA test for 13 oncogenic HPV types. HC2 relies on the formation of target HPV DNA-RNA probe heteroduplexes during the hybridization step in specimens that are positive for ≥ 1 oncogenic HPV types (HPV type 16 [HPV16], HPV18, HPV31, HPV33, HPV35, HPV39, HPV45, HPV51, HPV52, HPV56, HPV58, HPV59, and HPV68). Detection relies on chemiluminescence of these hybrids by using an alkaline phosphatase-conjugated monoclonal antibody specific to DNA-RNA complexes with dioxetane substrate in a 96-well enzyme-linked immunosorbent assay format. After liquid-based, ThinPrep cytology slides (Cytyc Corporation, Boxborough, MA) were prepared, 4-mL aliquots of the residual in the PreservCyt vials were used for HPV DNA testing by HC2. Signal strengths in relative light units (RLU) were compared with 1 pg/mL HPV16 DNA-positive controls (RLU/PC). The Food and Drug Administration-approved 1.0 RLU/PC ($\approx$ 1 pg/mL) as the threshold for a positive result.[31] Of the 5060 women enrolled into ALTS, we had valid HC2 results on 4819 women (95.2%).

We also used L1 consensus primer PGMY09/11 polymerase chain reaction (PCR) amplification and reverse-line blot hybridization for type-specific detection[32] on cervical specimens collected into specimen transport medium (STM; Digene Corporation, Gaithersburg, MD) from each patient. Specimens were thawed, and a 150-μL aliquot was digested by adding 7.5 μL of digestion solution (20 mg/mL proteinase K, 10% laureth-12, 20 mM Tris, and 1 mM ethylenediamine tetraacetic acid [EDTA], pH 8.5) and incubating at 60 °C for 1 hour. DNA from the digested material was precipitated by adding 1.0 mL of absolute ethanol containing 0.5 M ammonium acetate, incubating the mixture overnight at − 20 °C, and centrifuging for 30 minutes at $\times$ 13,000 *g*. The supernatant was discarded immediately, and the crude DNA pellet was dried overnight at room temperature. The pellet was resuspended in 50 μL of 20 mM Tris and 1 mM EDTA, pH 8.5.

We amplified 5 μL of each sample by using the PGMY09/11 L1 consensus primer system and AmpliTaq gold polymerase (Perkin Elmer, Wellesley, MA). Amplifications were done in a thermal cycler (model 9600; Perkin Elmer) using the following algorithm: 9-minute AmpliTaq gold activation at 95 °C followed by 40 cycles of 1-minute denaturation at 95 °C, 1-minute annealing at 55 °C, and 1-minute extension at 72 °C, and a 5-minute final extension at 72 °C.

Reverse-line blotting using HPV genotyping strips (Roche Molecular Systems, Alameda, CA) was used to detect 27 HPV genotypes (HPV6, HPV11, HPV16, HPV18, HPV26, HPV31, HPV33, HPV35, HPV39, HPV40, HPV42, HPV45, HPV51, HPV52, HPV53, HPV54, HPV55, HPV56, HPV57, HPV58, HPV59, HPV66, HPV68, HPV73 [PAP238A], HPV82 subtype [W13B], HPV83 [PAP291], and HPV84 [PAP155]) and a β-globin internal control. For 3000 women, we tested for 11 additional nononcogenic genotypes (HPV61, HPV62, HPV64, HPV67, HPV69, HPV70, HPV71, HPV72, HPV81, HPV82 subtype [IS39], HPV89

[CP6108]). Of the 5060 women enrolled into ALTS, we had valid PCR tests on 4915 women (97.1%).

HPV Classification

Using both HC2 and PCR data, we classified HPV DNA status as positive or negative for oncogenic types[33]: oncogenic HPV-positive if HC2 or PCR results were positive for HPV16, HPV18, HPV31, HPV33, HPV35, HPV39, HPV45, HPV51, HPV52, HPV56, HPV58, HPV59, or HPV68; otherwise, HPV DNA status was classified as negative for oncogenic HPV. Among the women who had negative results for oncogenic HPV, we reclassified as nononcogenic HPV-positive those women who had a positive PCR result for any HPV type other than the 13 oncogenic types listed above. We conservatively reclassified women (n = 202) as having a nononcogenic HPV type if they were HC2 positive but PCR negative for oncogenic types and positive for HPV6, HPV53, HPV66, HPV67, HPV70 and/or HPV81, recognizing that HC2 occasionally cross-reacts with these types, especially in cervical specimens from women with cytologic abnormalities.[34] Of the 5060 women enrolled into ALTS, 5052 women (99.8%) had at least 1 test result, and 4682 women (92.5%) had both tests; women who had only 1 HPV test result were classified accordingly using the results available.

Pathology

Clinical management was based on the clinical center pathologists' cytologic and histologic diagnoses. In addition, all referral smears, ThinPreps, and histology slides were sent to the Pathology QC Group (QC Pathology) based at Johns Hopkins University Medical Center for review and secondary diagnoses.

Our outcome of interest was defined as ≥ CIN3, including histologic CIN3 and the very few women (n = 7 patients) who had carcinoma that was detected cumulatively either at enrollment or during the 2-year follow-up, as diagnosed by the QC Pathology review. We used this rigorous definition in recognition that CIN3 detected within 2 years of an HPV DNA positive test is more likely to be a missed prevalent case than a true incident case, because a single colposcopic evaluation with biopsy and histologic evaluation is not perfectly sensitive for the detection of CIN3 and carcinoma,[27] and CIN3 rarely develops from an HPV infection within 2 years. In contrast, cervical intraepithelial neoplasia Grade 2 (CIN2) is a poorly reproducible diagnosis[35] that may represent an admixture of CIN1 and CIN3. Therefore, we included CIN2 in the multivariate models (described below) as an intermediate outcome that was excluded from the primary case definition (CIN3, including the few carcinomas) and from controls (women with oncogenic HPV and ≤ CIN2), thereby creating a conceptual "buffer zone" between infection and CIN3. In this analysis, which was restricted to women with oncogenic HPV (n = 3133 women), 506 of 542 women (93.4%) with ≥ CIN3 and 361 of 397 women (90.9%) with CIN2 diagnosed in ALTS were included, demonstrating the extraordinarily strong correlation between oncogenic HPV detection and diagnoses of ≥ CIN2 (i.e., only 7.7% of ≥ CIN2 detected over 2 years were HPV DNA-negative at enrollment).

Statistical Analyses

Univariate logistic regression was conducted to identify the variables that were associated with positive oncogenic HPV status in women without disease (i.e., < CIN2) compared with HPV-negative women without disease. All variables that were associated significantly with oncogenic HPV positivity in univariate analyses were included in a multivariate model. Final models were adjusted for age (18–19 years, 20–24 years, 25–29 years, 30–34 years, and ≥ 35 years), recent (in the past year) numbers of sexual partners (0, 1, ≥ 2), and lifetime numbers of sexual partners (0–2, ≥ 3). Only nine women reported no sexual activity. We examined the interaction of lifetime and recent number of sexual partners by examining the effect of lifetime number of partners in strata defined by recent number of partners. Odds ratios (ORs) and 95% confidence intervals (95%CIs) were calculated.

To determine the risk for disease (CIN2, ≥ CIN3), we restricted these analyses to women who had an oncogenic HPV-positive infection at enrollment; we also included the factors identified above as predictors of HPV positivity. ORs and 95%CIs adjusted for relevant parameters (e.g., predictors of oncogenic HPV, established risk factors for cervical neoplasia), were determined with multinomial logistic regression modeling for CIN2 and ≥ CIN3 compared with controls (< CIN2). Our final model examining the associations between the 3 SES indicators (education, race, and source of medical care) and CIN2 and ≥ CIN3, was adjusted for the following variables: age, smoking history, number of Papanicolaou (Pap) smears in the last 5 years, parity, referral diagnosis, and study center. OC use, history of vulvar warts, condom use, and difficulty becoming pregnant were not informative in the model and, thus, were excluded. A dose-response correlation for education (P for trend) was assessed in the models by treating the education variable as continuous (which assumes a linear trend).

TABLE 1
Predictors of Oncogenic Human Papillomavirus (HPV) Positivity: Distribution among HPV-Negative, Nononcogenic HPV-Positive, and Oncogenic HPV-Positive Women[a]

	HPV negative (n = 1343)[b]		Non-oncogenic HPV positive (n = 504)[b]		Oncogenic HPV positive (n = 2266)[b]		
Variable	No.	%	No.	%	No.	%	OR (95% CI)
Study center							
Alabama	324	24.1	151	30.0	767	33.8	1.0 (ref)
Oklahoma	213	15.9	91	18.1	425	18.8	0.9 (0.7–1.1)
Pennsylvania	444	33.1	86	17.1	364	16.1	0.5 (0.4–0.7)[c]
Washington	362	27.0	176	34.9	710	31.3	0.7 (0.5–0.8)[c]
Referral diagnosis							
ASCUS	1180	87.9	367	72.8	1397	61.7	1.0 (ref)
LSIL	163	12.1	137	27.2	869	38.3	3.4 (2.8–4.2)[c]
Age at enrollment							
18–19 yrs	81	6.0	64	12.7	352	15.5	1.0 (ref)
20–24 yrs	257	19.1	186	36.9	973	42.9	0.8 (0.6–1.1)
25–29 yrs	258	19.2	112	22.2	503	22.2	0.4 (0.3–0.6)[c]
30–34 yrs	190	14.1	50	9.9	225	9.9	0.3 (0.2–0.4)[c]
≥ 35 yrs	557	41.5	92	18.3	213	9.4	0.1 (0.1–0.1)[c]
Recent/lifetime no. of sexual partners							
0/0–2	46	3.5	4	0.8	13	0.6	1.0 (ref)
0/3+	60	4.5	23	4.6	32	1.4	2.2 (1.0–5.0)
1/0–2	320	24.1	67	13.4	266	11.8	1.6 (0.8–3.2)
1/3+	682	51.4	240	48.0	1149	51.1	3.5 (1.8–6.7)[c]
2+/0–2	9	0.7	8	1.6	28	1.2	3.9 (1.4–11.0)[c]
2+/3+	211	15.9	158	31.6	759	33.8	5.5 (2.8–10.9)[c]
Smoking history							
Never	749	55.8	268	53.3	1256	55.6	1.0 (ref)
Former	239	17.8	70	13.9	195	8.6	0.6 (0.5–0.8)[c]
Current	355	26.4	165	32.8	810	35.8	1.1 (0.9–1.3)
Oral contraceptive use							
No/never used birth control	697	52.2	196	39.1	814	36.1	1.0 (ref)
Use in past 2 yrs, not current	136	10.2	96	19.2	425	18.9	1.4 (1.1–1.7)[c]
Current user	502	37.6	209	41.7	1015	45.0	0.9 (0.8–1.1)
History of vulvar warts							
No	1206	90.1	436	86.9	1938	85.9	1.0 (ref)
Yes, treated	110	8.2	50	10.0	228	10.1	1.1 (0.9–1.5)
Yes, untreated	22	1.6	16	3.2	90	4.0	1.8 (1.1–3.0)[c]
Condom use							
No/never used birth control	625	46.7	146	29.2	511	22.7	1.0 (ref)
Use in past 2 yrs, not current	408	30.5	173	34.6	902	40.0	1.2 (1.0–1.4)
Current user	305	22.8	181	36.2	841	37.3	1.4 (1.1–1.7)[c]
Difficulty becoming pregnant							
Never tried or had difficulty	1270	94.7	491	97.4	2228	98.5	1.0 (ref)
Yes	71	5.3	13	2.6	35	1.5	0.5 (0.3–0.8)[c]

HPV: human papillomavirus; OR: odds ratio; 95%CI: 95% confidence interval; ref: reference; ASCUS: atypical squamous cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesion.

[a] ORs with 95%CIs from a multivariate logistic regression model comparing oncogenic HPV-positive women with HPV-negative women. The analysis was restricted to women who had a diagnosis of cervical intraepithelial neoplasia less than Grade 2 during the 2-year study period (i.e., non-cases). Adjusted for all variables in table.

[b] Total numbers may not add up due to missing data.

[c] For these ORs, the lower or upper confidence bound does not include 1.00.

RESULTS

Oncogenic HPV-positive women without disease (i.e., < CIN2) differed significantly from the HPV-negative women without disease by several characteristics. SES indicators and medical, gynecologic, contraceptive, reproductive, sexual, and smoking history variables were associated significantly with positive oncogenic HPV status in univariate, unadjusted analyses (data not shown). After accounting for age and sexual behavior (recent and lifetime number of sexual partners)

TABLE 2
Association of Socioeconomic Status Indicators with Oncogenic Human Papillomavirus Positivity

	OR (95%CI)					
Indicators of SES	**Unadjusted**	**Adjusted[a]**	**Adjusted[b]**	**Adjusted[c]**	**Adjusted[d]**	**Adjusted[e]**
Education						
Completed college/grad school	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Some college/vocational school	1.7 (1.4–2.1)[f]	1.4 (1.2–1.7)[f]	1.0 (0.8–1.3)	1.0 (0.8–1.3)	1.0 (0.8–1.2)	1.0 (0.8–1.2)
Completed high school	2.0 (1.6–2.4)[f]	1.5 (1.2–1.8)[f]	1.2 (0.9–1.5)	1.2 (1.0–1.6)	1.1 (0.9–1.4)	1.1 (0.8–1.4)
Less than high school	2.3 (1.8–2.9)[f]	1.5 (1.2–2.0)[f]	1.1 (0.8–1.5)	1.1 (0.8–1.6)	1.0 (0.7–1.3)	0.9 (0.7–1.3)
Race/ethnicity						
White/non-Hispanic	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Black	1.6 (1.4–1.9)[f]	1.3 (1.1–1.6)[f]	1.3 (1.1–1.6)[f]	1.3 (1.1–1.6)[f]	1.2 (1.0–1.5)	1.2 (0.9–1.4)
White/Hispanic	1.4 (1.0–2.1)	1.2 (0.8–1.8)	1.0 (0.7–1.5)	1.1 (0.7–1.7)	1.1 (0.7–1.7)	1.1 (0.7–1.7)
Asian/Pacific Islander	1.1 (0.8–1.6)	1.1 (0.7–1.6)	0.8 (0.5–1.2)	0.8 (0.5–1.3)	0.8 (0.5–1.3)	0.9 (0.6–1.3)
American Indian/Alaskan Native	1.6 (1.0–2.6)[f]	1.4 (0.9–2.3)	1.2 (0.7–2.1)	1.2 (0.7–2.1)	1.1 (0.6–1.9)	1.1 (0.6–1.9)
Source of medical care						
Insurance	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Self-pay/partner or family-pay	2.1 (1.8–2.4)[f]	1.9 (1.6–2.2)[f]	1.4 (1.2–1.7)[f]	1.3 (1.1–1.6)[f]	1.2 (0.9–1.4)	1.1 (0.9–1.4)
Medicaid/Medicare/other gov't	2.2 (1.8–2.7)[f]	1.8 (1.5–2.2)[f]	1.4 (1.1–1.8)[f]	1.3 (1.0–1.7)[f]	1.3 (1.0–1.7)[f]	1.3 (1.0–1.6)
No source	2.1 (1.4–3.1)[f]	1.6 (1.0–2.4)[f]	1.3 (0.8–2.1)	1.1 (0.7–1.8)	0.9 (0.6–1.5)	0.9 (0.5–1.4)

OR: odds ratio; 95%CI: 95% confidence interval; SES: socioeconomic status; ref: reference; gov't: government.

[a] Adjusted for SES indicators (education, race/ethnicity, source of medical care).

[b] Adjusted for SES indicators and age at enrollment (ages 18–19 years, 20–24 years, 25–29 years, 30–34 years, and ≥ 35 years).

[c] Adjusted for SES indicators, age at enrollment, and sexual behavior (recent and lifetime number of sexual partners in the following categories: 0 recent and 0–2 lifetime, 0 recent and ≥ 3 lifetime, 1 recent and 1–2 lifetime, 1 recent and ≥ 3 lifetime, 2 recent and 2 lifetime, ≥ 2 recent and ≥ 3 lifetime).

[d] Adjusted for SES indicators, age at enrollment, sexual behavior, and study center (Alabama, Oklahoma, Pennsylvania, and Washington).

[e] Adjusted for SES indicators, age at enrollment, sexual behavior, study center, and smoking history (never, former, current).

[f] For these ORs, the lower confidence bound does not include 1.00.

in multivariate models, most associations no longer were significant. Those factors that remained associated significantly or marginally with oncogenic HPV positivity in the multivariate models are shown in Table 1. In addition to the already established HPV risk factors of age and sexual behavior, we found that former OC use, former and current condom use, and history of untreated vulvar warts were associated with an increased risk for positive oncogenic HPV status. Characteristics that demonstrated a decreased risk for positive oncogenic HPV status included former smoking and difficulty becoming pregnant. Risk factors for infection with nononcogenic HPV were similar to those for infection with oncogenic HPV, although the associations were weaker (data not shown).

The SES indicators of race/ethnicity, education, and source of medical care all were associated with positive oncogenic HPV status in univariate, unadjusted models (Table 2). However, adjustment for confounding factors apparently explained these associations. After mutually adjusting for education, race/ethnicity, and source of medical care, the associations of the SES indicators with positive oncogenic HPV status weakened but remained significant (Table 2). After adjusting further for the main predictors of HPV infection—age and sexual behavior—only black women and women with government assistance or who paid for their own health care remained at increased risk for oncogenic HPV positivity. Black women were no longer at increased risk after stratification by study center. The associations of government health assistance or self-paid health care with positive oncogenic HPV status no longer were significant after smoking history was included in the multivariate model. One possible exception to the null findings was observed in subgroup analyses that were stratified by study center: In those analyses, white/Hispanic women in Oklahoma had an increased risk of oncogenic HPV positivity (OR, 2.7; 95%CI, 1.2–6.5) compared with white/non-Hispanic women. This may highlight the varying make-up of "Hispanic" ethnicity in different regions of the United States.

Among women who were positive for an oncogenic HPV type(s) at enrollment, SES indicators showed significant associations with a diagnosis of CIN2 or ≥ CIN3 (Table 3). Adjusting for the other SES indicators, age at enrollment, study center, referral diagnosis, smoking history, number of Pap smears in past years, and parity, women with less than a high school education, completion of high school, or com-

TABLE 3
Association of Socioeconomic Status Indicators with Cervical Intraepithelial Neoplasia Grade 2 and Grade ≥ 3 among Oncogenic Human Papillomavirus DNA-Positive Women[a]

	Total		< CIN2		CIN2[b]			> CIN3[b]		
SES indicators	No.	%	No.	%	No.	%	OR[c] (95%CI)	No.	%	OR[c] (95%CI)
Education										
Completed college	392	12.5	306	13.5	40	11.1	1.0 (ref)	46	9.1	1.0 (ref)
Some college	1175	37.5	853	37.7	140	38.8	1.2 (0.8–1.8)	182	36.0	1.5 (1.1–2.2)[d]
Completed high school	1002	32.0	745	32.9	105	29.1	1.0 (0.6–1.6)	152	30.0	1.5 (1.0–2.3)[d]
Less than high school	561	17.9	359	15.9	76	21.1	1.4 (0.8–2.3)	126	24.9	2.4 (1.5–3.7)[d,e]
Race/ethnicity										
White/non-Hispanic	1814	58.2	1240	54.9	219	60.8	1.0 (ref)	355	70.9	1.0 (ref)
Black	1015	32.5	798	35.3	106	29.4	0.8 (0.5–1.0)	111	22.2	0.5 (0.4–0.7)[d]
White/Hispanic	115	3.7	90	4.0	15	4.2	0.9 (0.5–1.7)	10	2.0	0.4 (0.2–0.8)[d]
Asian/Pacific Islander	100	3.2	72	3.2	11	3.1	0.9 (0.5–1.8)	17	3.4	1.0 (0.5–1.7)
American Indian/Alaskan Native	75	2.4	58	2.6	9	2.5	0.9 (0.5–2.0)	8	1.6	0.5 (0.2–1.1)
Source of medical care										
Insurance	639	20.5	467	20.7	77	21.3	1.0 (ref)	95	18.8	1.0 (ref)
Self-pay	1654	53.0	1160	51.4	198	54.8	0.9 (0.7–1.3)	296	58.6	1.1 (0.8–1.4)
Medicaid/Medicare/other gov't assistance	729	23.3	556	24.6	72	19.9	0.7 (0.5–1.0)[d]	101	20.0	0.8 (0.5–1.1)
No source	101	3.2	74	3.3	14	3.9	1.1 (0.6–2.2)	13	2.6	0.8 (0.4–1.6)

< CIN2: cervical intraepithelial neoplasia less than Grade 2; OR: odds ratio; 95%CI: 95% confidence interval; SES: socioeconomic status; ref: reference; gov't: government.

[a] ORs with 95%CIs were determined from multinomial logistic regression models comparing women who had a CIN2 or ≥ CIN3 diagnosis with women who had a < CIN2 diagnosis. The analysis included all women who were oncogenic human papillomavirus-positive at enrollment, i.e., women with all diagnoses (< CIN2, CIN2, ≥ CIN3) during the 2-year study period.

[b] Includes all patients who were diagnosed at enrollment, during the 2-year follow-up, and at the exit colposcopy.

[c] Adjusted for other SES indicators, age at enrollment, study center, referral diagnosis/study arm, smoking history, number of Papanicolaou smears in past 5 years prior to enrollment, and parity.

[d] For these ORs, the lower or upper confidence bound does not include 1.00.

[e] *P* value for trend = 0.0035.

pletion of some college had an increased risk for a ≥ CIN3 outcome compared with women who had completed college: OR, 2.4 (95%CI, 1.5–3.7); OR, 1.5 (95%CI, 1.0–2.3); and OR, 1.5 (95%CI, 1.1–2.2), respectively. A trend of increasing risk for ≥ CIN3 with decreasing education level was evident (*P* for trend = 0.0035). Within strata defined by race/ethnicity, the increased risks associated with education levels less than completion of college were present for black, white/non-Hispanic, and Asian women.

Among women who were positive for oncogenic HPV, black women and white/Hispanic women had a decreased risk for ≥ CIN3 compared with white/non-Hispanic women (OR, 0.5; 95%CI, 0.4–0.7; OR, 0.4; 95%CI, 0.2–0.8, respectively). This pattern was evident in all educational strata. The decreased risks for ≥ CIN3 in black women and white/Hispanic women were particularly striking in Oklahoma (OR, 0.1; 95%CI, 0.0–0.5; OR, 0.2; 95%CI, 0.0–0.7, respectively). Asian/Pacific Islander women (OR, 1.0; 95%CI, 0.5–1.7) did not have significantly different risk for ≥ CIN3 compared with white/non-Hispanic women. There were small numbers of American Indian/Alaskan Native women in ALTS (OR, 0.5; 95%CI, 0.2–1.1).

The source of medical care was not associated significantly with a diagnosis of ≥ CIN3. None of the SES indicators had a significant association with a diagnosis of CIN2 at any point during the 2-year follow-up. Combining SES variables (e.g., black women with less education and medical coverage) did not reveal any associations with ≥ CIN3.

DISCUSSION

The current study provides evidence that an indicator of lower SES—fewer years of education—is associated with a higher risk for ≥ CIN3, even after controlling for screening, follow-up, and other major risk factors for oncogenic HPV infection. First, we examined predictors of oncogenic HPV positivity in a group of women who did not develop the main outcome of ≥ CIN3 or the intermediate outcome of CIN2 during the course of the 2-year trial, because women who develop cervical neoplasia are less likely to represent the general population at risk for oncogenic HPV infection. In this population of women who were referred with cervical cytologic abnormalities, as expected, the prevalence of HPV infection was higher in the ALTS study than the prevalence found in the general population.[36] The predictors of oncogenic HPV infection in ALTS were consistent with previous studies, which demonstrated

that young age and increased numbers of sexual partners were associated with an increased risk for oncogenic HPV infection.[37] Although the literature is inconsistent regarding the associations of smoking and OC use with oncogenic HPV infection, we found that former smoking was associated with a decreased risk for oncogenic HPV, and former OC use was associated with an increased risk for oncogenic HPV. In addition, we found that women who consulted a clinician about difficulty becoming pregnant were less likely to have oncogenic HPV, an association that remained significant even after adjusting for age, sexual behavior, and study center. We believe that this predictor most likely is linked to unmeasured sociodemographic and behavioral factors rather than a biologic phenomenon.

Initially, we found that the SES indicators of education, race/ethnicity, and source of medical care were associated with HPV infection; however, these associations diminished once age, sexual behavior, study center, and smoking history were taken into account. Other studies that have found a preponderance of HPV in low socioeconomic groups observed this high prevalence in association with younger age and greater number of sexual partners.[38–40] Thus, high HPV prevalence appears to be the result of behaviors, not SES per se.

Although we believe that ALTS provided excellent data to study the risks of cervical neoplasia associated with SES in which follow-up and treatment were equalized, our correlates of SES were not optimal. SES is measured best by a combination of indicators that include income, education, occupation, census tract, and living situation, among other factors. Also, our results may not be generalizable to unscreened women, because all of the women in the ALTS trial were referred due to an abnormal screening test.

Nonetheless, restricting our analyses to women who were infected with oncogenic HPV allowed us to focus on the risks associated with potential HPV cofactors. In our total of 506 women with ≥ CIN3, only 7 women had carcinoma, and the rest had CIN3. Among the 7 women who had a diagnosis of carcinoma, 2 women were black, and 5 women were white/non-Hispanic; 1 woman had less than a high school education, 2 women had completed high school, 3 women had completed some college, and 1 woman had completed college; and 4 women had medical insurance, whereas 3 women self-paid for their medical care. A previous study reported higher rates of cervical precancer and lower rates of cervical carcinoma in the white population than in minority populations, possibly due to differential access to screening.[18] In ALTS, participants came from a screened population of women; they were referred to the trial because of an abnormal Pap smear. We observed that black and white/Hispanic women had a decreased risk for ≥ CIN3 compared with white/non-Hispanic women after controlling for oncogenic HPV positivity and screening/follow-up. This differs from the increased risk observed previously in black and white/Hispanic women; however, those studies did not account for HPV infection and screening.[17–19] Our finding needs to be replicated before we conclude that black and white/Hispanic women are at truly decreased risk of progression to ≥ CIN3 given equal screening/follow-up and HPV infection status. Only if this is confirmed would the reasons underlying decreased risk be worth pursuing.

The Institute of Medicine 2003 report *Unequal Treatment* reported that racial/ethnic health disparities still exist after controlling for insurance, follow-up, and severity of disease.[24] These differences may be due to biases on the part of both health professionals and patients.[41] In the ALTS randomized trial, all women were treated according to study protocol regardless of their race/ethnicity. Thus, our findings suggest that, in a setting in which screening and follow-up are equalized, the historically observed disparities seen in racial/ethnic minority groups would be minimized or, conceivably, reversed.

Our finding that less than a college education is associated with increased risk for ≥ CIN3, even in the ALTS setting of equalized follow-up and treatment, is of interest. We postulated that this association may be due to unequal access to care prior to entry into ALTS. To explore this idea, we performed a subanalysis separating women who received a ≥ CIN3 diagnosis at enrollment from women who received their diagnosis during follow-up or at exit. Although the differences were not significant, the trend of increasing risk of ≥ CIN3 with decreasing education indeed was stronger at enrollment (less than a high school education: OR, 2.8; completed high school: OR, 1.7; some college: OR, 1.7) than during the 2-year uniform ALTS protocol (less than high school education: OR, 1.8; completed high school: OR, 1.3; some college, OR, 1.3). Thus, there may be other factors we have not recognized that occur before recruitment that are associated with lower levels of education and increase the likelihood that a woman infected with oncogenic HPV will progress to ≥ CIN3. Some of these unknown factors may have included barriers to health care and not understanding the importance of follow-up. The risks associated with race/ethnicity and with the source of medical care did not differ greatly between women who were diagnosed with ≥ CIN3 at enrollment and women who were diagnosed during follow-up or at exit (data not shown).

In conclusion, the current results have shown that SES is not associated significantly with oncogenic HPV infection after accounting for other established risk factors for HPV. However, educational levels less than completion of college appear to be associated with ≥ CIN3 given oncogenic HPV infection. Despite historic racial and ethnic disparities, women of black and white/Hispanic race/ethnicity appeared to be at decreased risk for ≥ CIN3 compared with women of white/non-Hispanic race/ethnicity in a setting in which screening, treatment, and follow-up were equivalent. Thus, ongoing efforts should emphasize methods for equalizing screening and follow-up among women of varying SES, regardless of race or ethnicity.

REFERENCES

1. Parkin DM, Pisani P, Ferlay J. Global cancer statistics. *CA Cancer J Clin.* 1999;49:33–64.
2. Moreno V, Bosch FX, Munoz N, et al. Effect of oral contraceptives on risk of cervical cancer in women with human papillomavirus infection: the IARC multicentric case-control study. *Lancet.* 2002;359:1085–1092.
3. Castellsague X, Munoz N. Chapter 3: cofactors in human papillomavirus carcinogenesis—role of parity, oral contraceptives, and tobacco smoking. *J Natl Cancer Inst Monogr.* 2003;31:20–28.
4. Smith JS, Green J, Berrington DG, et al. Cervical cancer and use of hormonal contraceptives: a systematic review. *Lancet.* 2003;361:1159–1167.
5. Munoz N, Franceschi S, Bosetti C, et al. Role of parity and human papillomavirus in cervical cancer: the IARC multicentric case-control study. *Lancet.* 2002;359:1093–1101.
6. Deacon JM, Evans CD, Yule R, et al. Sexual behaviour and smoking as determinants of cervical HPV infection and of CIN3 among those infected: a case-control study nested within the Manchester cohort. *Br J Cancer.* 2000;83:1565–1572.
7. Castle PE, Wacholder S, Lorincz AT, et al. A prospective study of high-grade cervical neoplasia risk among human papillomavirus-infected women. *J Natl Cancer Inst.* 2002;94:1406–1414.
8. Plummer M, Herrero R, Franceschi S, et al. Smoking and cervical cancer: pooled analysis of the IARC multi-centric case-control study. *Cancer Causes Control.* 2003;14:805–814.
9. Wang SS, Hildesheim A. Chapter 5: viral and host factors in human papillomavirus persistence and progression. *J Natl Cancer Inst Monogr.* 2003;31:35–40.
10. Smith JS, Munoz N, Herrero R, et al. Evidence for Chlamydia trachomatis as a human papillomavirus cofactor in the etiology of invasive cervical cancer in Brazil and the Philippines. *J Infect Dis.* 2002;185:324–331.
11. Smith JS, Munoz N, Franceschi S, Eluf-Neto J, Herrero R, Peeling RW. Chlamydia trachomatis and cervical squamous cell carcinoma. *JAMA.* 2001;285:1704–1706.
12. Smith JS, Herrero R, Bosetti C, et al. Herpes simplex virus-2 as a human papillomavirus cofactor in the etiology of invasive cervical cancer. *J Natl Cancer Inst.* 2002;94:1604–1613.
13. Smith JS, Bosetti C, Munoz N, et al. Chlamydia trachomatis and invasive cervical cancer: a pooled analysis of the IARC multicentric case-control study. *Int J Cancer.* 2004;111:431–439.
14. Parikh S, Brennan P, Boffetta P. Meta-analysis of social inequality and the risk of cervical cancer. *Int J Cancer.* 2003;105:687–691.
15. Schiffman M, Hildesheim A. Cervical cancer. In: Schottenfeld D, Fraumeni JF, Jr., editors. Cancer epidemiology and prevention, 3rd ed. New York: Oxford University Press. In Press.
16. Jones CP. Invited commentary: "race," racism, and the practice of epidemiology. *Am J Epidemiol.* 2001;154:299–304.
17. Devesa SS, Diamond EL. Association of breast cancer and cervical cancer incidence with income and education among whites and blacks. *J Natl Cancer Inst.* 1980;65:515–528.
18. Wang SS, Sherman ME, Hildesheim A, Lacey JV Jr., Devesa S. Cervical adenocarcinoma and squamous cell carcinoma incidence trends among white women and black women in the United States for 1976–2000. *Cancer.* 2004;100:1035–1044.
19. O'Brien K, Cokkinides V, Jemal A, et al. Cancer statistics for Hispanics, 2003. *CA Cancer J Clin.* 2003;53:208–226.
20. Taylor VM, Jackson JC, Schwartz SM, Tu SP, Thompson B. Cervical cancer among Asian American women: a neglected health problem? *Asian Am Pac Isl J Health.* 1996;4:327–342.
21. Bach PB, Schrag D, Brawley OW, Galaznik A, Yakren S, Begg CB. Survival of blacks and whites after a cancer diagnosis. *JAMA.* 2002;287:2106–2113.
22. Brawley OW, Freeman HP. Race and outcomes: is this the end of the beginning for minority health research? *J Natl Cancer Inst.* 1999;91:1908–1909.
23. Schwartz KL, Crossley-May H, Vigneau FD, Brown K, Banerjee M. Race, socioeconomic status and stage at diagnosis for five common malignancies. *Cancer Causes Control.* 2003;14:761–766.
24. The Institute of Medicine. Unequal treatment; confronting racial and ethnic disparities in health care. Washington, DC: The National Academic Press, 2003.
25. Drain PK, Holmes KK, Hughes JP, Koutsky LA. Determinants of cervical cancer rates in developing countries. *Int J Cancer.* 2002;100:199–205.
26. Schiffman M, Adrianza ME. ASCUS-LSIL Triage Study. Design, methods and characteristics of trial participants. *Acta Cytol.* 2000;44:726–742.
27. Guido R, Schiffman M, Solomon D, Burke L. Postcolposcopy management strategies for women referred with low-grade squamous intraepithelial lesions or human papillomavirus DNA-positive atypical squamous cells of undetermined significance: a two-year prospective study. *Am J Obstet Gynecol.* 2003;188:1401–1495.
28. Cox JT, Schiffman M, Solomon D. Prospective follow-up suggests similar risk of subsequent cervical intraepithelial neoplasia Grade 2 or 3 among women with cervical intraepithelial neoplasia Grade 1 or negative colposcopy and directed biopsy. *Am J Obstet Gynecol.* 2003;188:1406–1412.
29. ASCUS-LSIL Triage Study (ALTS) Group. A randomized trial on the management of low-grade squamous intraepithelial lesion cytology interpretations. *Am J Obstet Gynecol.* 2003;188:1393–1400.
30. ASCUS-LSIL Triage Study (ALTS) Group. Results of a randomized trial on the management of cytology interpretations of atypical squamous cells of undetermined significance. *Am J Obstet Gynecol.* 2003;188:1383–1392.

31. Schiffman M, Herrero R, Hildesheim A, et al. HPV DNA testing in cervical cancer screening: results from women in a high-risk province of Costa Rica. *JAMA.* 2000;283:87–93.
32. Gravitt PE, Peyton CL, Alessi TQ, et al. Improved amplification of genital human papillomaviruses. *J Clin Microbiol.* 2000;38:357–361.
33. Bosch FX, Manos MM, Munoz N, et al. Prevalence of human papillomavirus in cervical cancer: a worldwide perspective. International Biological Study on Cervical Cancer (IBSCC) Study Group. *J Natl Cancer Inst.* 1995;87:796–802.
34. Castle PE, Schiffman M, Burk RD, et al. Restricted cross-reactivity of hybrid capture 2 with nononcogenic human papillomavirus types. *Cancer Epidemiol Biomarkers Prev.* 2002;11:1394–1139.
35. Stoler MH, Schiffman M. Interobserver reproducibility of cervical cytologic and histologic interpretations: realistic estimates from the ASCUS-LSIL Triage Study. *JAMA.* 2001;285: 1500–1505.
36. Sherman ME, Lorincz AT, Scott DR, et al. Baseline cytology, human papillomavirus testing, and risk for cervical neoplasia: a 10-year cohort analysis. *J Natl Cancer Inst.* 2003;95: 46–52.
37. Schiffman M, Kjaer SK. Chapter 2: Natural history of anogenital human papillomavirus infection and neoplasia. *J Natl Cancer Inst Monogr.* 2003;31:14–19.
38. Bauer HM, Hildesheim A, Schiffman MH, et al. Determinants of genital human papillomavirus infection in low-risk women in Portland, Oregon. *Sex Transm Dis.* 1993;20:274–278.
39. Hildesheim A, Gravitt P, Schiffman MH, et al. Determinants of genital human papillomavirus infection in low-income women in Washington, DC. *Sex Transm Dis.* 1993;20:279–285.
40. Wheeler CM, Parmenter CA, Hunt WC, et al. Determinants of genital human papillomavirus infection among cytologically normal women attending the University of New Mexico student health center. *Sex Transm Dis.* 1993;20:286–289.
41. Bach PB, Pham HH, Schrag D, Tate RC, Hargraves JL. Primary care physicians who treat blacks and whites. *N Engl J Med.* 2004;351:575–584.